Biological Forum – An International Journal **13(3a): 639-642(2021)**

ISSN No. (Print): 0975-1130
ISSN No. (Online): 2249-3239

Potential Utilization of Liquorice Extract as Natural Preservative for Tuna Fish during Chilled Storage

Aditya Singh*, S.P. Patel and A.R. Dodia
Department of Fish Processing Technology,
College of Fisheries Science, Kamdhenu University, Veraval, (Gujarat), India.

(Corresponding author: Aditya Singh)*
(Received 26 July 2021, Accepted 29 September, 2021)
(Published by Research Trend, Website: www.researchtrend.net)

ABSTRACT: Liquorice or Mulethi/Jethimadh/Yastimadhu scientifically known as *Glycyrrhiza glabra* is the most commonly used medicinal plant. It is popular within pharmaceutical industry as well as in household for its anti-inflammatory, anti-microbial and anti-tyrosinase activity. All these properties of liquorice are contribution of its different constituents. Availability of Liquorice extract is major challenge that we overcame. Out of which Glycyrrhizin which is anti-oxidant in nature has been used to prevent oxidation in tuna fish. Out of three solutions the tuna chunks dipped in 5% and 10% solutions depicted more freshness when measured with Torrymeter by the end of 9 days compared to control sample. Sensory evaluation indicated that both dipped samples outperformed control sample.

Keywords: Liquorice, Anti-inflammatory, Anti-microbial, Anti-tyrosinase, Glycyrrhizin, Oxidation, Torrymeter

INTRODUCTION

The importance of seafood is growing significantly all over the world due to its various beneficial health components which largely constitutes of protein and fats. Fats like omega-3 fatty acids are highly important for maintaining a healthy heart and body (Von Schacky and Harris 2007). They also control cholesterol and blood pressure. There is a sharp increase in demand of fish and fish-based products but there is also an unavoidable aspect that is spoilage of fish due to oxidation of fat (Lakshmanan *et al.,* 2002).

Lipids are oxidated by molecular oxygen into free radicals and is accelerated by heat, light and metal ions. Peroxides may further react with lipid forming new kind of peroxides. Second stage of lipid oxidation that is propagation phase persists until the final termination stage when two radicals combine. Due to these problems that occur post-harvest the seafood industry has started to look out for solutions out of which one of the best and most promising ones are antioxidants. Some of the antioxidants which are heavily used are Butylated hydroxy anisole (BHA), Butylated hydroxytoluene (BHT), octyl gallate (OG), propyl gallate (PG) and others.

Nowadays various researchers have started to experiment with antioxidants found in different natural herbs and fruits like tea, spinach, red wine etc. These natural oxidants from plant materials mainly consist of polyphenols (phenolic acid, flavonoids, anthocyanins), carotenoids and vitamins. There are various researches which demonstrate application of herbal extract (like tulsi) to fish chunks and their preserving effect (Suyani *et al.,* 2020). Liquorice roots (*Glycyrrhiza glabra*) belong to Mediterranean region, central to southern Russia and Asia. Liquorice roots are used in India from ancient times and part of daily routine. Food and Drug Administration (FDA) has approved liquorice root oil and is used in various products like beverages, toothpaste, chewing gums and cosmetics. *G. glabra* contains several chemical constituents like saponin, flavonoids, isoflavonoids, stilbenoids and coumarins. Glycyrrhiza is antioxidant in nature which will help in increasing in freshness of fish and provide longevity (Li *et al.,* 2011). It also acts as antiviral, anti-inflammatory & antioxidant (Mamedov and Egamberdieva 2019)

MATERIALS AND METHODS

Sample preparation. This study was held at College of Fisheries Science, Kamdhenu University, Veraval, Gujarat, India in the year 2021. To begin with, 2 kg Frigate tuna (*Auxisthazard*) was purchased from Kharakuwa fish market located near Veraval, Gujarat, India. Then washed and cut into chunks and again washed with tap water and divided into three groups (each group contains 180.04 ± 1.87g of chunks) and immediately placed on ice to cool before being treated with liquorice extract solutions. Tuna chunks divided into three groups were soaked in control (distilled water), 5% solution and 10% solution of liquorice extract and were named T1, T2 and T3. Then samples were taken out after 30 mins soaking and packed in LDPE plastic pouch. Control and liquorice extract treated samples were subjected to chilling for different intervals of time up to 9 days. Samples of control, 5% and 10% were analysed at 2 days interval for examination.

Analytical methods: Frigate tuna chunk samples were analysed from day 1 to 9 days at 2 days interval of chilled storage for their freshness, pH, colour and sensory evaluation. At each time of interval, the samples were thawed at room temperature prior to analysis. Freshness was measured using Torrymeter, used all over world as objective measurement of

freshness of fish caught in wide range of conditions. Electrode strip is stick to surface of tuna chunks, then take readings three times for control sample and meter will give us average freshness of the sample. To analyse the colour, the instrument colorimeter is used to measure L (lightness), a (redness and greenness), b (yellowness\blueness). Sensory evaluation was based on organoleptic test by scoring appearance, odour, colour and overall acceptability of treated and non-treated tuna chunk samples. The evaluation of tuna chunk samples was done by 9-point hedonic scale when 1, extremely dislike; 2, very much dislike; 3, moderately dislike; 4, slightly dislike; 5, neither like nor dislike; 6, slightly like; 7, moderately like; 8, very much like; 9, extremely like (Meilgaard, *et al*., 1999). pH was measured using pH meter.

Statistical analysis. All the statistical analysis was done in triplicates and data obtained were compared and analysed under Microsoft excel version 2019 software.

RESULT AND DISCUSSION

Freshness (using Torrymeter). Torrymeter is device used popularly for measuring freshness of fish.

Table 1: Mean freshness value of control (T1), T2 (5%) and T3 (10%).

Day	T1	T2	T3
0	7.9	7.6	7.7
1	8.0	8.2	8.0
3	6.7	8.7	8.7
5	5.9	7.3	8.8
7	5.7	6.1	7.7
9	4.7	5.7	7

It is robust, fully portable and suited for any use within fish processing industry and markets, processing factories or quality control laboratories, Freshness of the fish is indicated over the LCD display. The meter can be set to measure from 0 to 16 (Solanki *et al.*, 2016). This device works on menu driven software that sum the readings of samples and display their average value. Meter is most accurate in this mode. This device does not mark or damage the fish or sample either (Ravishankar *et al*., 1994). According to data in Table 1 T3 fish sample saw increase in freshness and lasted much longer compared to other two samples. Indicating that 10 % solution of liquorice extract enhanced freshness of tuna chunks to higher level than 5 % and distilled water.

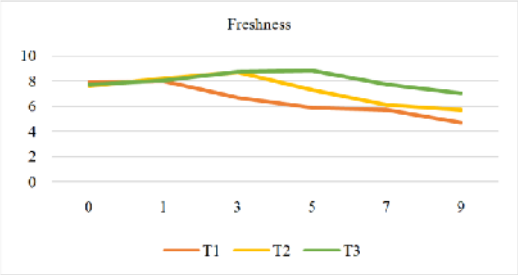

Fig. 1. Pictorial representation of mean freshness value.

Colour (using whiteness meter). The color of foods has been measured using L*a*b* or CIE Lab colour space, which is an international standard accepted by the Commission Internationale de l'Eclairage (CIE) in 1976 (Dhimmar *et al.*, 2019). Colour profile was measured by whiteness meter (Konica Minolta Colorimeter CR-14) which has L*, a* and b* values. L* denotes lightness, a* (redness) and b* (yellowness) values were recorded on tuna fish chunks of T1, T2 and T3 in the Petri plate (Fofandi Durga, *et al.*, 2019). Reading were taken for Day 0 (Before treatment) and Day 1 to 9 (after treatment). Data depicts that L (Lightness) value is high for T3 sample indicating that it is lightest among three sample. As for (Redness) a-value T1 has the lowest value on 9th day i.e., colour of this sample is somewhat greenish as value is decreasing. Whereas other two samples T2 and T3 are having increase in a-value. Yellowness or b-value is decreasing for T1 sample. Whereas increasing for T2 and T3. This may be due to brownish-yellowish colour of tincture.

Table 2: L* a* b* values before treatment.

Day 0	C	5%	10%
L	35.4 ± 0.50	37.7 ± 0.2	33.3 ± 0.42
a	-1.9 ± 0.25	-2.2 ± 0.31	-1.8 ± 0.21
b	+5.6 ± 0.26	+6.1 ± 0.36	+6.3 ± 0.3

Table 3: L* a* b* values after treatment.

Days	T1(C)	T2	T3
1	L = 39± 0.15	L=42.2±0.21	L=43.6±0.42
	a = -2.3± 0.35	a=-2.7±0.15	a=-2.4±0.11
	b = +9.7±0.2	b=+9.2±0.30	b=+10.5±0.2
3	L = 37.4±0.44	L=42.8±0.1	L=44.3±0.35
	a = -2.7±0.26	a=-2.5±0.15	a=-1.9±0.15
	b = +9.6±0.15	b=+10.6±0.26	b=+11.8±0.26
5	L = 36±0.26	L=43.3±0.45	L=44.7±0.38
	a = -3.6±0.32	a=-2.7±0.15	a=-1.4±0.2
	b = +9±0.25	b=+11±0.3	b=+12.6±0.2
7	L = 36.2±0.36	L=44.4±0.40	L=46.5±0.50
	a = -4.6±0.30	a=-1.6±0.15	a=-1.0±0.15
	b = +8.6±0.25	b=+11.4±0.21	b=+13.5±0.21
9	L = 35.2±0.56	L=45.4±0.46	L=48±0.06
	a = -5.5±0.15	a= -1.5±0.15	a= -1.2±0.36
	b =+7.8±0.36	b=+12.5±0.44	b=+14.7±0.17

Sensory evaluation: The sensory evaluation was performed by 5-member semi-trained panellist (Peryam and Pilgrim 1957). The panellist evaluated the colour, appearance, odour and overall acceptability of treated fish based on a nine-point hedonic scale. (AOAC, 2000)

The outer appearance of the treated fish seemed to be slightly different than control due to brown colour of extract. The results indicated that both T2 and T3 samples odour was slightly better than control. Ultimately T3 ousted T2 and T1 sample.

Table 4: Scores for sensory evaluation.

Storage period (Days)	Chunk samples	Appearance	Colour	Odour	Overall acceptability
0 (Before treatment)	T1 (Control)	8.7 ± 0.58	8.7 ± 0.58	9 ± 0	8.3 ± 1.15
	T2	8.3 ± 0.58	8.6 ± 0.58	9 ±0	8.7 ± 0.58
	T3	8 ± 1	8.4 ± 0.58	9±0	8.4 ± 0.58
1	T1 (Control)	8.3 ± 0.58	7.7± 0.58	6.7 ± 0.58	7.3 ± 1.15
	T2	7.7 ± 0.58	8 ± 0	7.4 ± 0.58	8.3 ± 0.58
	T3	8.3 ± 0.58	7.3 ± 0.58	8.7 ± 0.58	8.3 ± 1.15
3	T1 (Control)	6.7 ± 0.58	6.6 ± 0.58	5.7 ± 0.58	6.7 ± 0.58
	T2	7.3 ± 0.58	6.7 ± 0.58	7 ± 1	7.7 ± 0.58
	T3	8 ± 0	6.6 ± 1.53	8.3 ± 1.15	7.7 ± 0.58
5	T1 (Control)	6.4 ± 0.58	6 ± 0	5.4 ± 0.58	5.4 ± 0.58
	T2	6.4 ± 0.56	6.4 ± 0.58	6.7 ± 0.58	6.6 ± 1.53
	T3	6.7 ± 0.58	6 ± 1.73	7.3 ± 0.58	7.3 ± 0.58
7	T1 (Control)	4 ± 1	5 ± 1	4.7 ± 0.58	4.7 ± 0.58
	T2	5.7 ± 0.58	5.7 ± 0.58	5.6 ± 0.58	5.4 ± 0.58
	T3	6.3 ± 0.58	5.4 ± 0.58	7.3 ± 1.15	6.4 ± 1.15
9	T1 (Control)	3.3 ± 0.58	4 ± 1	3.3 ± 0.58	3.6 ± 0.58
	T2	4 ± 1	5 ± 1	4.7 ± 0.58	4.3 ± 0.58
	T3	6 ± 1	4.7 ± 1.15	6.4 ± 0.58	5.7 ± 0.58

pH value. pH is measured to evaluate whether fish has become too much acidic or not. To measure pH, we used pH meter. From control sample (T1) a small piece of flesh is taken. It is homogenised using distilled water in beaker. Then the probe of pH machine is dipped into three solutions with fixed pH to calibrate the probe. After that probe is washed again with distilled water and then dipped in homogenised mixture of flesh and distilled water. Once the reading gets stable note down the pH. Data for pH is provided in Table 5. The water holding capacity of muscles decreases with decrease in pH (Swatland, 2002). Thus, fall in pH indicates spoilage.

Table 5: pH values for fish samples.

Day	T_1	T_2	T_3
0	6.4 ± 0.06	6.43± 0.06	6.4 ± 0.1
1	6.4 ± 0.12	6.67± 0.12	6.7±0.1
3	6.3 ± 0.1	6.7± 0.1	6.83 ± 0.06
5	6.37 ± 0.12	6.63± 0.15	6.7 ± 0.1
7	6.13 ± 0.06	6.47± 0.12	6.57 ± 0.06
9	6 ± 0.17	6.23± 0.15	6.37 ± 0.58

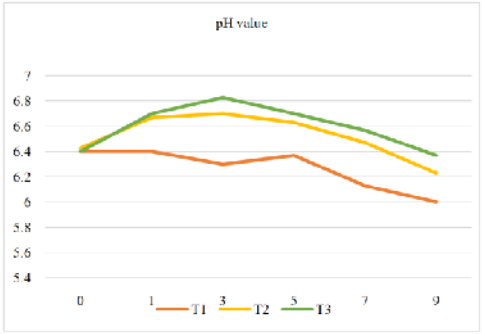

Fig. 2. Pictorial representation of mean pH value.

CONCLUSION

In this research the effect of herbal extract made from *Glycyrrhiza glabra* on tuna fish was investigated. There was no significant difference in rate of weight loss but slight difference in pH was observed where T3 was slightly basic as compared to T2 and T1. Freshness increased after treatment and was much higher in treated samples T3 and T2 compared to T1. Liquorice extract increased the lightness of fish (T3), whereas it's decreased in control sample (T1). There is slight colour change in fish due to dark colour of tincture but it compensates for the bearable odour of treated samples. Overall, at the end of 8th day sample T3 was more acceptable than T2. Then T2 is much more ahead of control in terms of acceptability.

This experiment showed that extract prepared out of liquorice (*Glycyrrhiza glabra*) expanded the freshness of fish when compared to control. More refining of extract could have been done in order to reduce its effect on colour of fish flesh. But overall, the result suggests that there is vast amount of research associated with Liquorice which can be done in near future. Every other property of liquorice root can be exploited to advance in fisheries.

Acknowledgement. We would like to thank Department of Fish Processing Technology for aiding our research work financially and also would like to thank all the faculty members who lend their support.
Conflict of Interest. None.

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How to cite this article: Singh, A., Patel, S.P. and Dodia, A.R. (2021). Potential Utilization of Liquorice Extract as Natural Preservative for Tuna Fish during Chilled Storage. *Biological Forum – An International Journal*, *13*(3a): 639-642.